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The Study of Correlation Between Heavy Metal Levels in Environment and Autism Case in Samarinda and Bantul

Studi Hubungan Antara Kadar Logam Berat di Lingkungan dengan Kejadian Autisme di Samarinda dan Bantul

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ABSTRACT
Autism is a disorder of pervasive development in children, which until now, the exact cause is unknown. The strongest allegations are genetic and environmental factors, especially heavy metals. This study aims to look the relationship between the occurrence of heavy metals pollution with the autism cases in Samarinda and Bantul. This study was carried out by measuring the levels of heavy metals in fish, vegetables, water and soil and also hair of 30 autistic children and 10 control children in Samarinda and 30 autism 4 control in Bantul.All samples were measured their heavy metal content (As, Hg, Cd, Pb) at The Central Nuclear Applied Science and Technology Laboratory in Bandung. The result compares with the standard value and were found that the samples was exceeding the heavy metals value in Samarinda is Arsenic in fish, lead in fish. While in Bantul, Mercury in fish, mercury in vegetable and lead in vegetable. In the hair samples result, the level of arsenic, mercury and lead were higher in autism children compared to controls in Samarinda, while in Bantul only mercury and lead levels are higher in hair of autism children than controls. Cadmium levelswere no difference between autistic and control hair both in Samarinda and Bantul.

ABSTRAK
Autisme adalah gangguan pervasive pada anak yang hingga saat ini penyebabnya yang pasti belum diketahui. Dugaan sangat kuat, penyebabnya adalah faktor genetik dan lingkungan hidup, khususnya oleh logam berat. Penelitian ini bertujuan untuk melihat adanya hubungan antara terjadinya paparan kadar logam berat di lingkungan dengan terjadinya penyakit autisme. Penelitian ini dilakukan dengan mengambil sampel ikan, sayuran, air dan tanah serta sampel rambut dari 30 orang anak autisme dan 10 orang rambut kontrol di Samarinda dan 30 orang anak autisme dan 4 orang kontrol di Bantul, dimana semua sampel ini diukur kadar logam beratnya (As, Hg, Cd, Pb) di laboratorium Pusat Sains dan Teknologi Nuklir Terapan Bandung. Hasil penelitian dilakukan tabulasi dan dibandingkan dengan standard. Hasilnya bahwa pada sampel lingkungan di Samarinda didapatkan bahwa kadar logam berat yang melebihi standar adalah timbal pada ikan (Lempake). Sedangkan di Bantul yang tinggi kadarnya adalah merkuri pada semua sampel ikan dan sayur, serta timbal pada sampel sayur dari Banguntapan. Pada rambut, baik di Samarinda maupun Bantul, kadar logam berat arsen, merkuri dan timbal lebih tinggi pada penderita autisme dibandingkan kontrol, sedangkan kadmium tidak ada perbedaan antara kelompok penderita dan kontrol.

INTRODUCTION

Currently in Indonesia, there has been a decline in environmental quality caused by environmental damage, both naturally occurring such as volcanic eruptions, earthquakes, as well as damage caused by human behavior such as industrialization, mining, agriculture which causes various pollution and one of them is pollutants that are very dangerous are heavy metals because they can cause health problems if they enter the human body.[1] The entry of heavy metals into the human body is mainly through food and drink, and through breathing. Symptoms that arise as a result of the entry of heavy metals into the human body depend on the reaction caused by the heavy metal to the body's organs.[2] The United States Department of Health gives main attention to three types of heavy metals that are considered the most dangerous, namely Arsenic (As), Lead (Pb) and Mercury (Hg) because these three chemicals can cause neurological disorders in the form of Attention Deficit Hyperactivity Disorders (ADHD), low IQ and Autism Spectrum Disorders (ASD) even though the levels are small.[3] The entry of heavy metals into the human body mainly through the food chain. If the consumption of foods containing heavy metals continues, it will cause bioaccumulation in the body and can cause systemic intoxication with disorders in the form of neurotoxic, nephrotoxic, fetotoxic and teratogenic effects. The disorder will be more severe if the body's detoxification ability decreases.[4]

Autism was chosen as the research subject because there were more cases and it was suspected that the influence of environmental factors (heavy metals) was very strong in the occurrence of this disease. The number of people with autism around the world is increasing, this is very concerning because most of the children suffering from autism are difficult to expect to become quality human resources. Based on data from the Autism Research Institute in 1987, in the United States only 1 person with autism was found in every 5000 children. Currently there is an increase, for every 110 children there is 1 person with autism.[5] Nationally, the number of people with autism is also increasing rapidly. In 1990 the number was only 2-5 cases/10,000 births, increasing to 20/10,000 births in 2010.[2] Meanwhile at the Kalimantan regional level, the province of East Kalimantan is a province with a high number of autism sufferers, allegedly related to the presence of industry and mining, particularly coal mining.

This research, which was conducted in Samarinda and Bantul, aims to see the influence of environmental factors, especially heavy metals. The city of Samarinda was chosen as the research location because in this city there are many coal mining activities and in the middle of this city there is the Mahakam river which is the place for new rock carriers from upstream to downstream (sea). Based on data from all special schools in Samarinda, the number of people with autism in this city is 197 people out of 812,597 residents. As a comparison, Bantul (Yogyakarta) district was chosen because in this district there is no large industry or coal mining area and based on data from the Ministry of Health's Basic Health Research (Riskesdas) in

2017, the number of people with autism in this area is not high.[6] Based on secondary data from all special schools in Bantul, in 2018 the number of autism sufferers was 97 out of 995,264 residents. This picture shows that the condition of environmental pollution is heavier in Samarinda than in Bantul, so cases of autism sufferers are also more prevalent in Samarinda.

This study aims to prove the existence of a relationship between exposure to heavy metals As, Hg, Cd and Pb in the environment and the occurrence of autism in Samarinda city and Bantul district. Specifically, this study aims to prove: 1) The presence of heavy metal levels in the environment (fish, vegetables, water and soil whose levels are higher than normal) (H1); 2) It proves that the heavy metal content in the environment in Samarinda is higher than in Bantul; 3) Proving that the levels of heavy metals in the hair of children with autism in Samarinda and Bantul were higher than normal and higher than controls (H1); 4) Proving that the levels of heavy metals in the hair of children with autism in Samarinda were higher than those of children with autism in Bantul (H1).

MATERIAL AND METHOD

This research is an exploratory research with a case control study design. The research was conducted in Samarinda city and Bantul Regency from September 18 to October 24 2018. The subjects used were hair from children with autism and hair from normal children (controls), as well as environmental samples. The number of respondents with autism children in Samarinda is 30 people, consisting of 20 boys and 10 girls. The youngest was 6 years 2 months old and the oldest was 16 years old. Control consisted of 10 people, 7 men and 3 women. The youngest was 6 years 5 months old and the oldest was 16 years old. Meanwhile, in Bantul, the number of children with autism was 30, 20 boys and 10 girls. The youngest was 5 years 2 months old and the oldest was 16 years old. Non autism controls totaled 4 people, consists of 3 men and 1 woman with the youngest age 9 years and the oldest 14 years 2 months. Respondents were selected by taking into account their domicile. It is expected that the available respondents will represent the entire research area. Likewise, the determination of respondents based on gender was adjusted to the ratio of the number of autism sufferers based on gender in both Samarinda and Bantul. Meanwhile, environmental samples taken and measured for heavy metal levels were fish, vegetables, water and soil. Fish samples were taken in Samarinda at four locations, namely in the Kelurahan Lempake (nile tilapia), Jembayan (nile tilapia), Sempaja (catfish), PPI Selili (catfish). The number of fish samples taken weighing 1 kg. Vegetable samples were taken from Lempake (mustard greens), Sempaja (swamp cabbage and mustard greens). Each vegetable weighs 1 kg. To keep vegetables and fish fresh, they are put in ziplock plastic and then put in a cooler box equipped with ice gel. Water samples were taken from WTP PDAM Lempake, WTP Bengkuring and WTP Cendana. Each sample was taken as much as 1 liter. Then put it in a plastic bottle and cool it to keep it durable.

Soil samples were taken from the areas of Sempaja, Lempake and Samarinda Seberang (around the Mahakam River). Soil weight taken at each sample location is 1 kg. After that the sample is put into ziplock plastic. On the outside of all environmental samples labeled the type of sample, the location of collection (coordinates). The same is the case in Samarinda, in Bantul the same sample was taken. Fish samples were taken from Banguntapan (nile tilapia), Jetis (nile tilapia) and Singosaren Imogiri (nile tilapia) areas. Vegetable samples were taken from the Bantul-Yogya border area (mustard greens), Banguntapan area (lembayung vegetable). Water samples were taken from three locations, namely well water in Banguntapan, well water at Jalan Parangtritis Km 12.5 and well water from Rojowinangun. Meanwhile, soil sampling was carried out in two areas, namely the Banguntapan area and the Parangtritis Km 12.3 area. The number of environmental samples taken, the way of packing and labeling were carried out as in the sampling in Samarinda.

After all the samples are ready (both hair samples and environmental samples), they will be transferred to the laboratory of the Center for Applied Nuclear Science and Technology, Jalan Tamansari No. 71 Bandung. In this laboratory, heavy metal levels were measured for all types of samples. The heavy metals measured are Arsenic (As), Mercury (Hg), Cadmium (Cd) and Lead (Pb). Measurements were made using the Atomic Absorption Spectrometry (AAS) procedure for measuring levels of cadmium and lead and Neutron Activation Analysis (NAA) for measuring arsenic and mercury levels. Measurement results are presented in tabular form and narrated. Furthermore, a statistical test was carried out using parametric hypothesis testing (t test) or non-parametric (Mann Whitney test) to prove whether the levels of heavy metals in the hair of children with autism were higher than that of normal children (control). The t test is carried out if the two data tested are normally distributed. If it does not normally distributed, use the Mann Whitney test. The normality test was performed using the Kolmogorov-Smirnov (KS) test statistic. The data is said to be normally distributed if the p-value is >0.05, and the data does not normally distributed if the p-value is <0.05. Because this research uses human subjects so this research is preceded by requesting a review of the ethics of the Research and Community Service Institute for Research Ethics Commission Involving Hu-man Subjects of IPB, the fan has obtained Ethical Approval Number: 169/IT3.KEPMSM-IPB/SK/2018 dated 11 April 2018.

RESULTS

The results of statistical analysis for Arsenic (As) levels in Samarinda with the Mann Whitney test showed that the mean arsenic levels in the hair of children with autism were higher than in control hair. Meanwhile, in Bantul, the arsenic levels in the hair of children with autism were the same as in control hair. This means that arsenic has effect on the occurrence of autism in Samarinda, while in Bantul it does not affect the occurrence of autism. Analysis of mercury levels in Samarinda using the Mann

Whitney test showed that the average mercury level in the hair of children with autism was higher than that of controls. Likewise in Bantul, mercury levels in the hair of children with autism were higher than in control hair. This means that mercury affects the occurrence of autism in Samarinda and Bantul. For levels of lead (Pb), with the t test showed that the average hair lead level in children with autism in Samarinda was higher than the control. The same thing happened in Bantul. This means, both in Samarinda and Bantul, Pb has an effect on the occurrence of autism. Meanwhile, for cadmium (Cd), the Mann Whitney test showed that in Samarinda and Bantul there was no difference in hair levels between autism and control children. That is, cadmium has no effect on the occurrence of autism.

Levels of arsenic (As) in Samarinda that exceed normal are from fish samples caught from the Lempake area (Table 1), namely 0.46 mg/kg (maximum limit of 0.25 mg/kg based on Regulation of the Head of BPOM Number 23 of 2017).[9] Likewise, the water samples that were also taken from this region, the arsenic level also reached the maximum contamination limit of 0.013 mg/kg (0.01 mg/kg maximum limit) (Table 3). The other environmental samples had arsenic levels lower than the contamination limit. Likewise, in Bantul none of the samples had arsenic levels reaching the maximum limit. From these results it can be seen that arsenic levels are only high (exceeding the maximum contamination limit) in fish taken from Lempake Samarinda and water taken from Lempake.

Even though the levels of mercury (Hg) in Samarinda did not reach the maximum limit, all fish samples contained mercury levels (Table 1). Meanwhile in Bantul, all three fish samples exceeded the maximum contamination limit. Likewise, all four vegetable samples had levels that exceeded the maximum contamination limit (Table 2). Meanwhile, none of the water and soil samples had levels exceeding the maximum contamination limit. These results prove that for mercury levels, the environment in Bantul is more polluted than in Samarinda. While the levels of cadmium (Cd), neither in Samarinda nor Bantul, had levels that no exceeded the maximum contamination limit. In other words, cadmium in Samarinda and Bantul is the same.

The level of Lead (Pb) was found to exceed the maximum limit of contamination in fish samples taken at Lempake Samarinda, namely 3.23 mg/kg (maximum limit 0.10 mg/kg) (Table 1). Whereas in Bantul, samples whose Pb levels exceeded the maximum limit of contamination were vegetable samples taken from Bangun-tapan, namely 0.40 mg/kg (maximum limit of 0.2 mg/kg) (Table 2). From this description, it can be seen that Samarinda and Bantul are both polluted by Lead (Pb), but in Samarinda it is thought to have originated from mining activities, while in Bantul it is thought that emissions from burning gasoline by motorized vehicles, or perhaps from home industries such as industry used battery smelting. Meanwhile, none of the soil samples taken in Samarinda or Bantul had levels exceeding the contamination limit according to US.EPA standards.[10]

Table 4 shows the results of measurements of heavy metal levels in autism hair and control hair in Samarinda. The highest arsenic level in autism patients was found in respondent number 29 (0.0251 mg/kg), the lowest was at number 24 (0.053 mg/kg) with an average of 0.129 mg/kg. Whereas for control hair, the highest was in respondent number 37 (0.131 mg/kg), the lowest was in number 33 (0.023 mg/kg) with an average of 0.058.

Table 1. Tabulation of the Results of Measurements of Heavy Metal Levels in Fish in Samarinda and Bantul

Sampling Location	Sample Type	Arsenic Levels (mg/kg)	Mercury Levels (mg/kg)	Cadmium Levels (mg/kg)	Lead Levels (mg/kg)
Samarinda (Lempake)	Nile Tilapia	0.46	0.0021	≤ 0.0008	**3.23**
Samarinda (Jembayan)	Nile Tilapia	0.044	0.0048	≤ 0.0008	≤ 0.0128
Samarinda (Sempaja)	Patin	0.062	0.0018	≤ 0.0008	≤ 0.0128
Samarinda (PPI Selili)	Snakehead Fish	≤ 0.003	0.005	≤ 0.0008	≤ 0.0128
Bantul (Banguntapan)	Nile Tilapia	0.013	**0.295**	≤ 0.0008	≤ 0.0128
Bantul (Jetis)	Nile Tilapia	0.026	**0.272**	≤ 0.0008	≤ 0.0128
Bantul (Singosaren)	Nile Tilapia	0.029	**0.415**	≤ 0.0008	0.045
Maximum Contamination Limit		**0.25**	**0.2**	**0.06**	**0.1**

Source: Primary Data, 2018

Table 2. Tabulation of the Results of Measurements of Heavy Metal Levels in Vegetables in Samarinda and Bantul

Sampling Location	Sample Type	Arsenic Levels (mg/kg)	Mercury Levels (mg/kg)	Cadnium Levels (mg/kg)	Lead Levels (mg/kg)
Samarinda (Lempake)	Mustard Greens	0.006	0.0006	≤ 0.0008	0.072
Samarinda (Sempaja)	Swamp Cabbage	0.007	0.0007	0.0015	0.0955
Samarinda (Sempaja)	Mustard Greens	0.006	0.0010	0.0013	≤ 0.0128
Bantul (Perb.Bantul-Yogya)	Mustard Greens	0.031	**0.1628**	0.0026	0.29
Bantul (Banguntapan)	Lembayung Vegetable	0.002	**0.248**	0.0011	**0.40**
Bantul (Jetis)	Lembayung Vegetable	0.005	**0.17**	≤ 0.0008	0.065
Bantul (Singosaren)	Lembayung Vegetable	≤ 0.003	**0.378**	≤ 0.0008	0.07
Maximum Contamination Limit		**0.15**	**0.03**	**0.05**	**0.2**

Source: Primary Data, 2018

Table 3. Tabulation of the Results of Measurements of Heavy Metal Levels in Water in Samarinda and Bantul

Sampling Location	Sample Type	Arsenic levels (mg/L)	Mercury Levels (mg/L)	Cadmium Levels (mg/L)	Lead Levels (mg/L)
Samarinda (WTP PDAM Lempake)	AW.1	0.013	0.0008	≤ 0.00004	≤ 0.0004
Samarinda (WTP PDAM Bengkuring, Sempaja)	AW.2	≤ 0.007	0.0008	≤ 0.00004	0.0015
Samarinda (WTP Cendana)	AW. 3	≤ 0.007	0.0008	≤ 0.00004	≤ 0.0004
Bantul (Banguntapan)	ABL. 1	≤ 0.007	0.0007	≤ 0.00004	0.002
Bantul (Jl. Parangtritis Km. 12.3)	ABL. 2	≤ 0.007	0.0008	0.00005	0.0009
Bantul (Pilahan)	ABL. 3	≤ 0.007	0.0007	0.00009	0.0012
Quality Standards		**0.01**	**0.001**	**0.003**	**0.01**

Source: Primary Data, 2018

Analysis of arsenic levels in the hair of children with autism and control hair in Samarinda, showed that with the normality test using the Kolmogrov Smirnov (KS) test, data on children with autism did not normally distributed, while non-autistic control data normally distributed. Because one of the data did not normally distributed, the Mann-Whitney non-parametric test was continued. Mann Whitney test results with α = 0.05, indicating that the mean arsenic levels in the hair of children with autism in Samarinda were higher than controls. In other words, arsenic affects the incidence of autism in Samarinda.

Whereas for respondents with autism in Bantul (table 5) the highest arsenic levels were found in respondent number 30 (2.19 mg/kg), the lowest was in respondent number 11 (0.013 mg/kg) with an average of 0.151 mg/kg. whereas in control hair, the highest was respondent number 31 (0.088 mg/kg), the lowest was respondent number 33 (0.013 mg/ kg) with an average of 0.013. To see the difference in the mean levels of arsenic in the hair of autism and non-autistic controls, first a normality test was performed with the KS test. From the KS test, it was found that the data from the autism's hair was not normally distributed (p<0.05), while the data from the control hair was normally distributed (p> 0.05). Because one of the data did not normally distributed, to test for differences in arsenic levels in these two groups, a Mann-Whitney test was performed. With α = 0.05, it shows that the arsenic in the hair of children with autism is the same as that of non-autistic children. This means that arsenic has no effect on the occurrence of autism in Bantul.

The highest levels of mercury (Hg) in the hair of people with autism in Samarinda were found in respondent number 17 (2.28 mg/kg). Lowest at number 12 (0.091 mg / kg) with an average of 0.777 mg/kg (Table 4). Whereas for control hair, the highest was respondent number 35 (0.315 mg/kg), the lowest was number 32 (0.133 mg/kg) with an average of 0.226 mg/kg. The normality test with the KS test statistics showed that the data on children with autism did not normally distributed, while the non-autism data normally distributed. The results of the

Mann Whitney test with α = 0.05 showed which means that the mercury levels in the hair of children with autism in Samarinda were higher than in control hair. This means that mercury affects the occurrence of autism in Samarinda.

Table 4. Tabulation of Measurement Results of Heavy Metal Levels in Hair of Autism Patients and Controls in Samarinda

No	Gender	Age (Years, Months)	Arsenic (As) (mg/kg)	Mercury (Hg) (mg/kg)	Cadmium (Cd) (mg/kg)	Lead (Pb) (mg/kg)
			Autism			
1	Woman	9	0.144	0.778	0.008	0.183
2	Male	7,1	0.067	0.362	0.007	0.171
3	Male	9,2	0.112	0.413	0.051	0.307
4	Woman	8	0.152	0.408	0.044	0.367
5	Woman	7,6	0.092	0.561	0.043	0.426
6	Male	14	0.103	0.575	0.030	0.276
7	Male	8,4	0.096	0.813	0.031	0.726
8	Male	11	0.097	0.653	0.007	0.214
9	Male	13	0.143	0.721	0.019	0.158
10	Woman	9	0.091	0.834	0.015	0.246
11	Male	12	0.137	0.314	0.043	0.732
12	Woman	15.1	0.072	0.092	0.007	0.359
13	Male	14.8	0.081	0.351	0.002	0.631
14	Male	6.5	0.121	0.279	0.007	0.532
15	Woman	6.2	0.091	0.831	0.009	0.751
16	Male	16	0.085	1,321	0.035	0.151
17	Male	13	0.079	2,280	0.037	0.253
18	Woman	12.5	0.185	0.417	0.034	0.873
19	Male	14.5	0.093	0.923	0.011	1,701
20	Male	13	0.189	0.431	0.081	0.952
21	Male	14	0.178	0.831	0.048	1,651
22	Woman	12	0.093	0.753	0.050	1,345
23	Male	15	0.187	1,131	0.080	0.913
24	Male	15.4	0.053	0.451	0.048	0.852
25	Woman	11	0.217	0.852	0.050	0.545
26	Male	12	0.194	1,473	0.080	1,753
27	Male	13	0.132	1,253	0.088	1,651
28	Woman	14	0.115	0.371	0.055	0.851
29	Male	15	0.251	0.431	0.047	0.371
30	Male	13	0.213	0.583	0.082	0.521
			Control			
31	Male	6.5	0.033	0.213	0.021	0.210
32	Woman	7,2	0.051	0.133	0.015	0.213
33	Woman	8.5	0.023	0.151	0.010	0.131
34	Male	8.3	0.045	0.183	0.024	0.150
35	Male	9	0.031	0.315	0.015	0.090
36	Woman	10.5	0.069	0.153	0.021	0.187
37	Male	10	0.131	0.183	0.027	0.137
38	Male	15.5	0.083	0.263	0.030	0.081
39	Male	16	0.091	0.314	0.017	0.193
40	Male	14	0.027	0.155	0.043	0.135

Source: Primary Data, 2018

The highest mercury levels in children with autism in Bantul were found in respondent number 5 (1.177 mg/kg), the lowest was at number 28 (0.015 mg/kg) with an average of 0.334 mg/kg. Whereas in control hair, the highest Hg level was found in respondent number 31 (0.303 mg/kg), the lowest was in number 32 (0.136) with an average of 0.214 mg/kg (Table 5). The KS normality test showed that the data from the hair of children with autism did not normally distributed so that the Mann Whitney test was used to see the average difference in mercury levels in autistic and non-autistic hair. With $\alpha = 0.05$, it shows that mercury levels in Bantul are higher in autistic hair than non-autistic children (control).

Cadmium (Cd) levels in the hair of children with autism in Samarinda were highest in respondent number 11 (0.243 mg/kg), the lowest was at number 13 (0.02 mg/kg) with an average of 0.038 mg/kg. In control children, the highest level was in respondent number 40 (0.043 mg/kg), and the lowest was in number 33 (0.010 mg/kg) with an average of 0.038 mg/kg (Table 4). Statistical tests such as those carried out on arsenic and mercury, found that cadmium levels in the hair of children with autism were not higher than non-autistic children. This is consistent with environmental samples in which none of the levels exceed the maximum limit.

Likewise, in Bantul the highest levels of cadmium in the hair of children with autism were found in respondent number 22 (0.141 mg/kg), the lowest was in number 1,7,12,15 (0.007 mg/kg) with an average of 0.037 mg/kg. Whereas the highest control hair was at number 32 (0.037 mg/kg), the lowest was at number 33 (0.007 mg/kg) with an average of 0.019 mg/kg (Table 5). Statistical tests showed that the average cadmium level in the hair of children with autism was not higher than in control hair. Likewise, in environmental samples, none of which levels exceed the maximum limit.

The highest lead level (Pb) in the hair of children with autism in Samarinda was in respondent number 26 (1.753 mg/kg), the lowest was number 16 (1.151 mg/kg) with an average of 0.682 mg/kg. Whereas in control children, the highest was in number 32 (0.213 mg/kg), the lowest was at number 35 (0.09 mg/kg) with an average of 0.153 mg/kg. With the KS normality test, it was found that the two data were normally distributed (both of them had a p-value > 0.05). Because the two data are normally distributed, to see the difference in the mean of the two data, the t-test statistic is used. The results showed that the average hair lead levels in children with autism in Samarinda were higher than controls.

In Bantul, the highest Pb levels in the hair of children with autism were found in respondent number 23 (2.879 mg/kg), the lowest was in number 15 (0.071 mg/kg) with an average of 0.634 mg/kg. In control hair, the highest was in respondent number 33 (0.194 mg/kg), the lowest was in number 34 (0.124 mg/kg) with an average of 0.171 mg/kg (Table 5). In the statistical test, it was also found that the average lead level in the hair of children with autism was greater than the control.

Table 5. Tabulation of Measurement Results of Heavy Metal Levels in Hair of Autism Patients in Bantul

No	Gender	Age (Year, Month	Arsenic (As) (mg/kg)	Mercury (Hg) (mg/kg)	Cadmium (Cd) (mg/kg)	Lead (Pb) (mg/kg)
			Autism			
1	Male	13.5	0.136	0.217	0.007	0.128
2	Male	7.5	0.058	0.359	0.040	0.382
3	Male	14	0.150	0.110	0.069	0.763
4	Woman	9,2	0.032	0.207	0.020	0.440
5	Male	8.1	0.060	1,177	0.090	0.584
6	Woman	8.7	0.034	0.204	0.026	1,713
7	Male	12.2	0.049	0.175	0.007	0.085
8	Male	9	0.029	0.221	0.007	0.224
9	Male	15.6	0.041	0.157	0.012	0.156
10	Woman	14	0.078	0.397	0.042	1,290
11	Woman	13.6	0.013	0.227	0.105	0.714
12	Male	16	0.052	1,167	0.007	0.808
13	Woman	15	0.051	0.267	0.015	1,408
14	Male	15.8	0.061	0.152	0.014	1,083
15	Woman	9	0.052	0.228	0.007	0.071
16	Male	9.5	0.050	0.374	0.048	0.255
17	Male	7,2	0.073	0.397	0.030	0.144
18	Male	11	0.103	0.500	0.037	0.320
19	Male	6.5	0.063	1,067	0.015	0.995
20	Woman	6	0.066	0.600	0.031	0.195
21	Male	15.2	0.097	0.157	0.032	0.663
22	Male	14	0.043	0.138	0.141	0.969
23	Male	16	0.039	0.187	0.090	2,879
24	Male	9,2	0.138	0.731	0.017	0.189
25	Woman	15	0.125	0.125	0.047	0.404
26	Woman	14	0.048	0.107	0.013	0.475
27	Woman	9,2	0.063	0.138	0.011	0.206
28	Male	5,2	0.417	0.015	0.050	0.563
29	Male	14.2	0.106	0.203	0.034	0.765
30	Male	10	2,191	0.185	0.050	0.135
			Control			
31	Male	14.2	0.088	0.303	0.026	0.191
32	Woman	12	0.083	0.136	0.037	0.174
33	Male	9	0.013	0.227	0.007	0.194
34	Male	11	0.067	0.189	0.007	0.124

Source: Primary Data, 2018

DISCUSSION

Based on the Regulation of the Head of BPOM Number 23 of 2017, arsenic levels (As) in fish that exceed the contamination limit are found in fish taken from the Lempake Samarinda Area (0.46 mg/kg).[7] It is suspected that this arsenic source comes from coal mines because Around the fishing grounds (less than 2 km) there are coal mining activities. Even in this fish sample, not only arsenic was high but also lead (Pb) (3.23 mg/kg). It is estimated that waste from the coal mine goes into fish farming along with rainwater. The statistical results show that H_0 is rejected, which means that the arsenic level in Samarinda is higher than the arsenic level in Bantul. If these fish are eaten by humans it can cause intoxication, especially if the body's detoxification abilities are decreased.[8] This requires special attention, because fish is a food ingredient that is consumed by humans. If

repeated consumption, there will be accumulation in the body and one day it can have an impact on degenerative diseases.[9] The brain is an organ that is very sensitive to arsenic poisoning because it can directly affect brain development.[10] The results of measuring arsenic levels in Samarinda show that arsenic affect the occurrence of autism. Because the alleged source of arsenic in Samarinda comes from a coal mine, the issuance of a permit for a coal mining business should be accompanied by an AMDAL so that it will not endanger the community.

The age factor also determines the high levels of arsenic in the hair. The older the child is, the higher the arsenic level in the hair tends to be. For example, in people with autism in Samarinda (Table 4), respondent number 1 (9 years) had an arsenic level of 0.144 mg/kg higher than respondent number 2 (7 years) an arsenic level of 0.067 mg/kg, even though the two respondents were siblings. This difference is presumably due to differences in the duration of contamination. Likewise, with the domicile factor. The closer to the source of contamination, the higher the arsenic content in the hair tends to be. In Samarinda, it can be seen that the closer the domicile is to the coal mine location, the arsenic content in the hair is higher, for example respondent number 3 (0.112), number 6 (0.103), number 18 (0.185) and number 19 (0.093 mg/kg). How does arsenic enter the body of a child, still requires further research whether the entry of arsenic through the mother during pregnancy and into the fetus through the placenta or children who consume foods containing arsenic. High levels of arsenic in children's hair come not only from food and drink, but also from environmental pollution (water and air).[11]

Comparison between arsenic levels in the hair of children with autism in Samarinda and Bantul, can be done using statistical calculations such as the time to calculate the difference between arsenic levels in the hair of patients and controls. The statistical results showed that H1 was accepted, which means that arsenic levels in the hair of children with autism were higher in Samarinda than in Bantul.

Mercury (Hg) whose levels exceed the maximum limit of contamination are all fish and vegetable samples in Bantul. It is suspected that the source comes from nature (volcanic eruptions) or from a power plant that uses coal power. All fish samples taken from Samarinda had mercury levels lower than the maximum contamination limit. However, because fish is a consumption material that is mostly eaten by humans, it still requires attention and caution because mercury can accumulate in the human body and cause adverse health effects. It can even experience biomagnification, which means that the levels in the human body are higher than the source.[12] If you compare the mercury levels in Bantul and Samarinda, indicates that H_0 is accepted, which means that mercury contamination in Bantul is higher than Samarinda. Likewise, if we compare the mercury levels in the hair of children with autism and controls in Bantul, it shows that the average levels of mercury in the hair of children with autism are

higher than the average hair of controls. This means that mercury affects the occurrence of autism in Bantul. The high levels of mercury in vegetables in Bantul, apart from being thought to have originated from volcanoes and power plants, could also be due to excessive pesticide spraying.

Mercury is a heavy metal that is toxic and has harmful effects on health. Several studies have shown that high levels of mercury in the blood are a risk factor for autism.[13] One of the ways that mercury can enter the body of children is vaccination because vaccines contain thimerosal (an organic form of mercury) which is used as an ingredient vaccine preservative. This is evident from the research of McKean, et al that autism sufferers are more likely to experience side effects after vaccination than normal children.[13] Mercury can also enter the body of children through breast milk. Filon, et al in his research proved that high levels of mercury in breast milk, will enter the body of a child with autism and can be detected in the child's hair.[14] As with arsenic, mercury also shows a link between age and the domicile where the child lives. In this case, the older the child is, the higher the Hg levels in the hair. Likewise, the closer to the source of contamination, the higher the Hg content in the hair. This is in accordance with the results of research from Hadjkacem, et al who found that mercury levels in the hair of children with autism were higher than controls and in younger children with autism had lower levels of mercury.[15] When compared to mercury levels in the hair of children with autism in Samarinda and Bantul, statistical analysis showed that H_0 was accepted. which means that mercury levels in the hair of children with autism in Bantul are higher than in Samarinda.

In measuring cadmium levels in the environment, neither in Samarinda nor in Bantul, none of the samples had levels exceeding the maximum limit. Likewise, in hair samples, both in Samarinda and Bantul, there were no cadmium levels that exceeded normal levels. Likewise, there was no difference between cadmium levels in the hair of children with autism and control hair. The inclusion of cadmium as one of the heavy metals studied is because in several previous studies found the effect of cadmium on the occurrence of autism, such as research by Akyuzlu, et al in Ankara Turkey, by examining the hair and urine of autism sufferers and controls, it was found that cadmium and lead levels were higher than normal.[16] Yurdakok, found that Cd, Pb and Hg were the heavy metals that most polluted the environment, but in contrast to the research conducted in Samarinda and Bontang, none of the environmental samples had levels of cadmium exceeding the maximum limit.[17] When compared between the average levels of cadmium in the hair of children with autism and cadmium in control hair, statistical analysis showed that H_0 was accepted, meaning that cadmium levels in the hair of children with autism in Samarinda were not different from levels of cadmium in autistic children in Bantul. Measurement of lead levels in the environment

in Samarinda only found high levels in fish samples in Lempake. Likewise, in Bantul, only one vegetable sample had high lead levels. High lead levels in fish were also found by Maddusa, et al on his research on the Tondano River. This is because the high levels of Pb in the water exceed the standard threshold according to the standards of the Ministry of Environment number 51 of 2004.[18] The increase in Pb concentrations in fish is influenced by the Pb concentration in the waters.

In the hair samples from respondents of children with autism in Samarinda and Bantul, both of them had higher lead levels than controls. This means that lead has an effect on the occurrence of autism in Samarinda and Bantul. According to Lee, et al, Lead affects the development of the nervous system, not only on the emergence of autism, but also with Attention Deficit Hyperactivity Disorder (ADHD).[19] Research conducted by the Illionis Department of Public Health showed that 300,000 children whose blood samples were taken, 2500 children it turns out to have lead levels ≥ 10 µg/dl.[20] Likewise research from Rahbar, et al which was also conducted in Jamaica showed that children whose homes live by the side of the highway, have higher levels of lead in their blood. This is related to the high level of lead from pollution caused by the combustion of fuel from motorized vehicles.[21] Pb exposure to pregnant women causes Pb to enter the body, and through the placenta the Pb enters the fetus.[22] One of the consequences of high lead levels in the blood, it will have an impact on the decrease in IQ in these children.[23] In this study, there was no measurement between the severity of autism and the levels of heavy metals. When compared between the lead levels in the hair of children with autism and controls, statistical analysis showed that H_1 was accepted, which means that the lead levels in the hair of children with autism in Samarinda were higher than the lead levels in the hair of children with autism in Bantul.

CONCLUSION AND RECOMMENDATION

Of the four types of heavy metals studied (As, Hg, Cd and Pb), it turns out that three of them, namely arsenic, mercury and lead have polluted the environment in Samarinda and Bantul. This is consistent with the results of measurements of heavy metal levels in the hair of children with autism and controls. Arsenic (As) levels in Samarinda were, on average, higher in hair of autism sufferers than in controls (Arsenic affects the occurrence of autism). Mercury (Hg) in Samarinda and Bantul, both of which have higher levels of Mercury than the control (an effect on the occurrence of autism). Cadmium levels in Samarinda and Bantul have no effect on the occurrence of autism because cadmium levels in the environment and in hair do not exceed normal limits. Whereas lead, both in Samarinda and Bantul have an influence on the occurrence of autism. The results of this study indicate that there has been heavy metal pollution in two research locations (Samarinda and Bantul) which have an impact on the occurrence of autism. In order to prevent this condition from getting worse in the future, it is recommended that the main company that will

carry out mining activities is obliged to carry out an AMDAL, and not to open a mine, mainly coal mining in the middle of a residential area, because coal mining will have a negative impact. against health. After the company operates, pay attention to environmental management and environmental monitoring. Likewise, for power plants that use coal energy, presumably pay attention to disposal of waste (dust) because it can pollute the environment, especially vegetables and fish.

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